

FIG. 1A

H36.D2.B7 Anti-Tissue Factor Light Chain Variable Region

GACATTCAGATGACCCAGTCTCCTGCCTCCCAGTCTGCATCTCTGGGAGAAAGTGTACCATCACATGC  
D I Q M T Q S P A S Q S A S L G E S V T I T C  
CTGGCAAGTCAGACCATTGATACATGGTTAGCATGGTATCAGCAGAAACCAGGGAAATCTCCTCAGCTC  
L A S Q T I D T W L A W Y Q Q K P G K S P Q L  
CTGATTTATGCTGCCACCAACTTGGCAGATGGGGTCCCATCAAGGTTTCAGTGGCAGTGGATCTGGCACA  
L I Y A A T N L A D G V P S R F S G S G S G T  
AAATTTTCTTTCAAGATCAGCAGCCTACAGGCTGAAGATTTTGTAATTATTACTGTCAACAAGTTTAC  
K F S F K I S S L Q A E D F V N Y Y C Q Q V Y  
AGTTCTCCATTACAGTTTCGGTGCTGGGACCAAGCTGGAGCTGAAA  
S S P F T F G A G T K L E L K

FIG. 1B

H36.D2.B7 Anti-Tissue Factor Heavy Chain Variable Region

GAGATCCAGCTGCAGCAGTCTGGACCTGAGCTGGTGAAGCCTGGGGCTTCAGTGCAGGTATCCTGCAAG  
E I Q L Q Q S G P E L V K P G A S V Q V S C K  
ACTTCTGGTTACTCATTCACTGACTACAACGTGTACTGGGTGAGGCAGAGCCATGGAAAGAGCCTTGAG  
T S G Y S F T D Y N V Y W V R Q S H G K S L E  
TGGATTGGATATATTGATCCTTACAATGGTATTACTATCTACGACCAGAACTTCAAGGGCAAGGCCACA  
W I G Y I D P Y N G I T I Y D Q N F K G K A T  
TTGACTGTTGACAAGTCTTCCACCACAGCCTTCATGCATCTCAACAGCCTGACATCTGACGACTCTGCA  
L T V D K S S T T A F M H L N S L T S D D S A  
GTTTATTTCTGTGCAAGAGATGTGACTACGGCCCTTGACTTCTGGGGCCAAGGCACCACTCTCACAGTC  
V Y F C A R D V T T A L D F W G Q G T T L T V  
TCCTCA  
S S

\* CDR regions underlined.

0990586-112101

Antibody	Apparent $K_d$ , $M^{-1}$	Apparent $K_d$ , M
By ELISA		
D2	$5.2 \times 10^9$	$1.9 \times 10^{-10}$
I47	$6.5 \times 10^9$	$1.5 \times 10^{-10}$
K73	$9.8 \times 10^9$	$1.0 \times 10^{-10}$
K80	$2.3 \times 10^9$	$4.3 \times 10^{-10}$
L102	$2.5 \times 10^9$	$4.0 \times 10^{-10}$
L133	$1.7 \times 10^9$	$5.9 \times 10^{-10}$
By BLACore		
<u>H36</u>	<u><math>3.1 \times 10^{10}</math></u>	<u><math>3.2 \times 10^{-11}</math></u>
I43	$2.3 \times 10^9$	<u><math>4.3 \times 10^{-10}</math></u>
I47	$3.2 \times 10^9$	<u><math>3.1 \times 10^{-10}</math></u>
L133	$4.6 \times 10^9$	<u><math>2.2 \times 10^{-10}</math></u>
M107	$1.1 \times 10^9$	<u><math>9.1 \times 10^{-10}</math></u>

FIG. 2

[illegible]

Antibody Name	% Inhibition Antibody Preincubated with TF/VIIa
D1	0
D1B	1
H31	4
<u>H36</u>	<u>95</u>
I43	1
J131	7
K80	0
K82	0
K87	1
L97B	7
L101	0
L102	0
L105	0
L133	0
M5	1
M107	34

**FIG. 3**

Antibody Name	<u>% Inhibition</u> TF Preincubated with Antibody Prior to Addition of VIIa	<u>% Inhibition</u> TF Preincubated with VIIa Prior to Addition of Antibody
D1	15	nd
D1B	48	12.7
H31	64	21
H36	0	0
I43	68	55
J131	38	11
K80	12	nd
K82	0	nd
K87	0	nd
L96	0	nd
L101	38	11
L102	14	nd
L105	4	nd
L133	13	nd
M5	0	nd
M107	0	nd

FIG. 4

[rhTF], nM	[H36.D2], nM	H36.D2/rhTF Molar Ratio	Clotting Time (seconds)	% Inhibition of rhTF Function
0.0048	0	0	102.3	0
	1.61	335.4	114.3	31.3
	3.23	670.8	121.3	45.8
0.023	0	0	77.6	0
	1.61	70.0	85.3	52.2
	3.23	140.0	91.1	65.2
	6.45	280.4	99.6	73.9
0.092	0	0	49.3	0
	3.23	35.1	65.8	65.2
	6.45	70.1	88.5	90.2
	12.90	140.2	113.3	95.7
0.46	0	0	32.6	0
	6.45	14.0	52.7	82.4
	12.90	28.0	80.2	96.7
	32.30	70.2	117.9	99.3
2.30	0	0	23.9	0
	16.10	7.0	47.1	94.4
	32.30	14.0	95.2	99.7
	64.50	28.0	115.3	99.9
11.52	0	0	22.2	0
	16.10	1.4	30.2	93.4
	32.30	2.8	46.0	98.8
	64.50	5.6	87.6	99.9
	161.30	14.0	114.0	100.0

FIG. 5

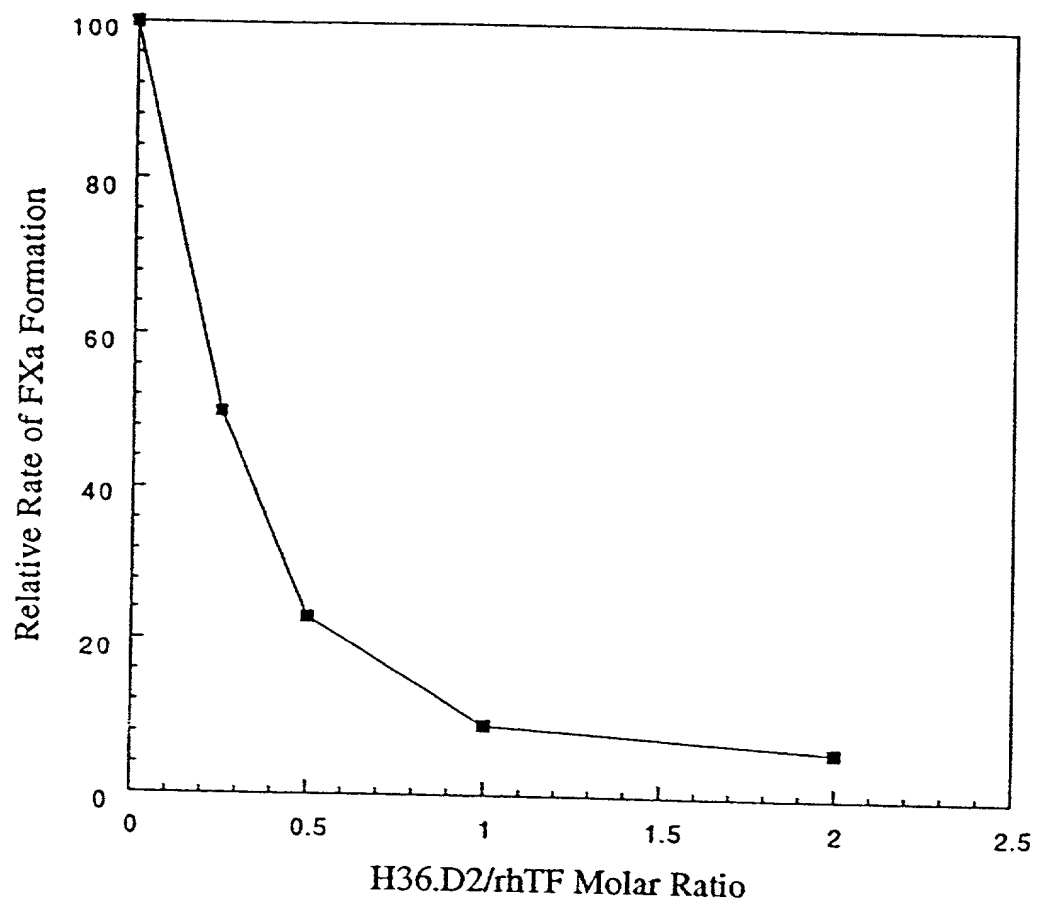


FIG. 6A

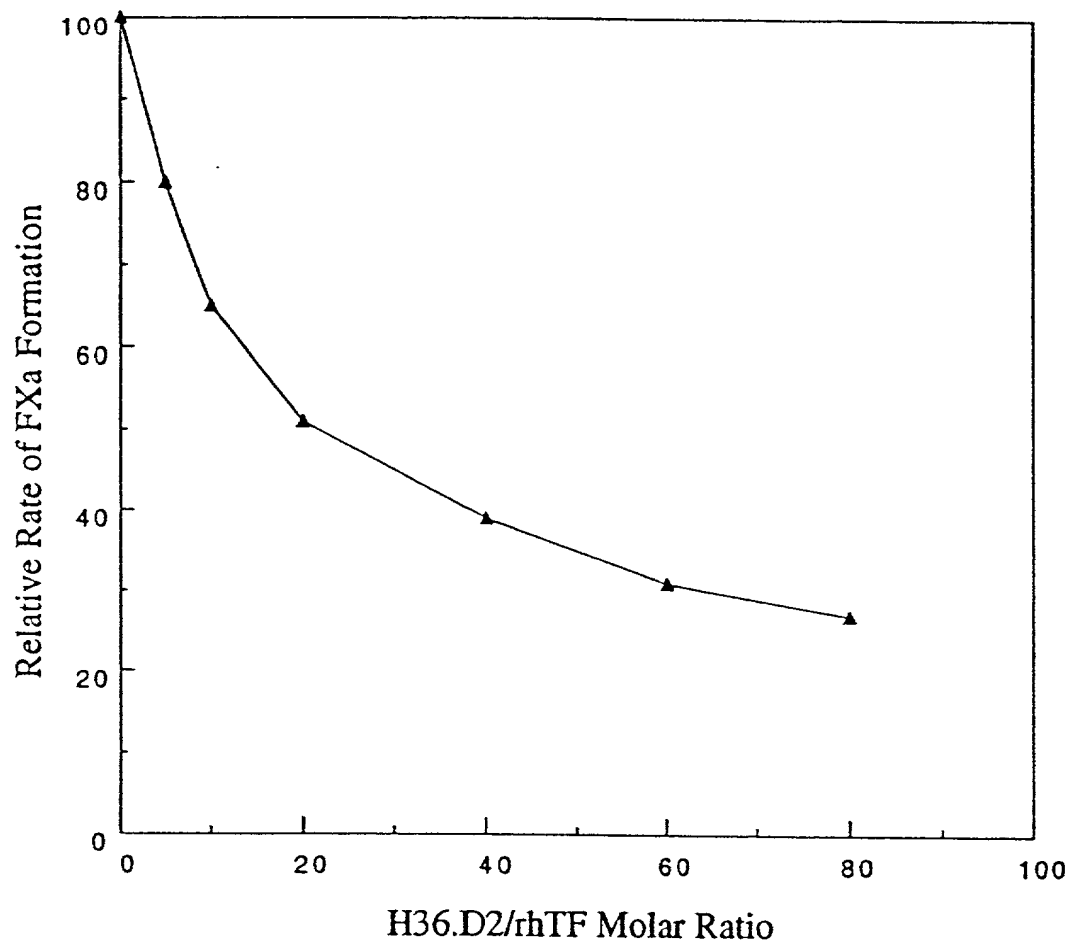


FIG. 6B

H36.D2 Concentration (ng)	<u>% Inhibition</u> Cells (TF/FVII) and H36.D2 preincubated prior to FX addition	<u>% Inhibition</u> FX and H36.D2 are added simultaneously to Cells (TF/FVII)
0	0	0
50	88	nd
100	92	nd
200	97	nd
800	nd	76
1600	nd	78
3200	nd	92

FIG. 7



TTTTT-99906669

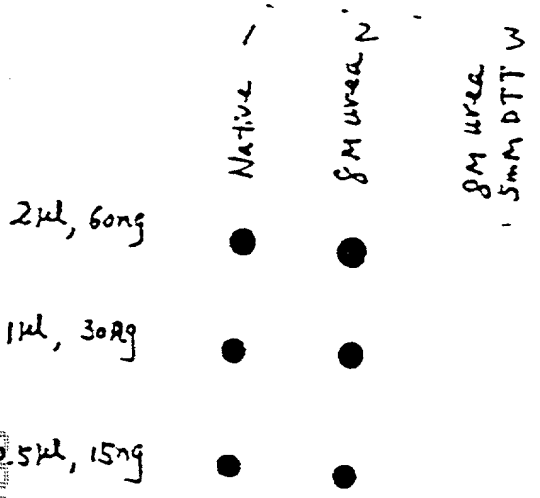


FIG. 8A

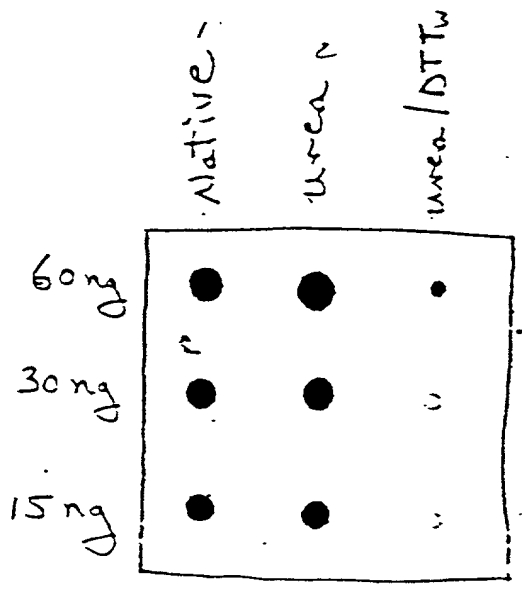
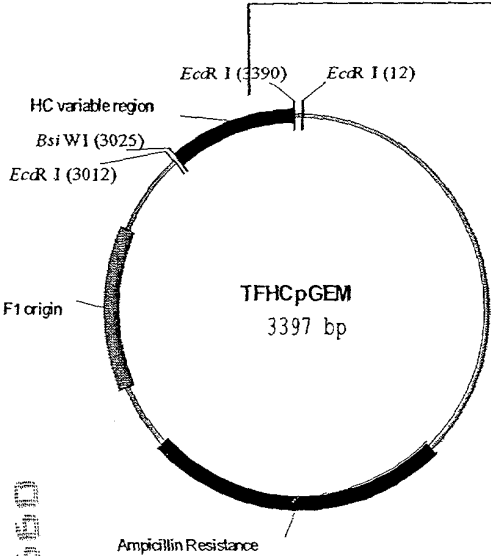


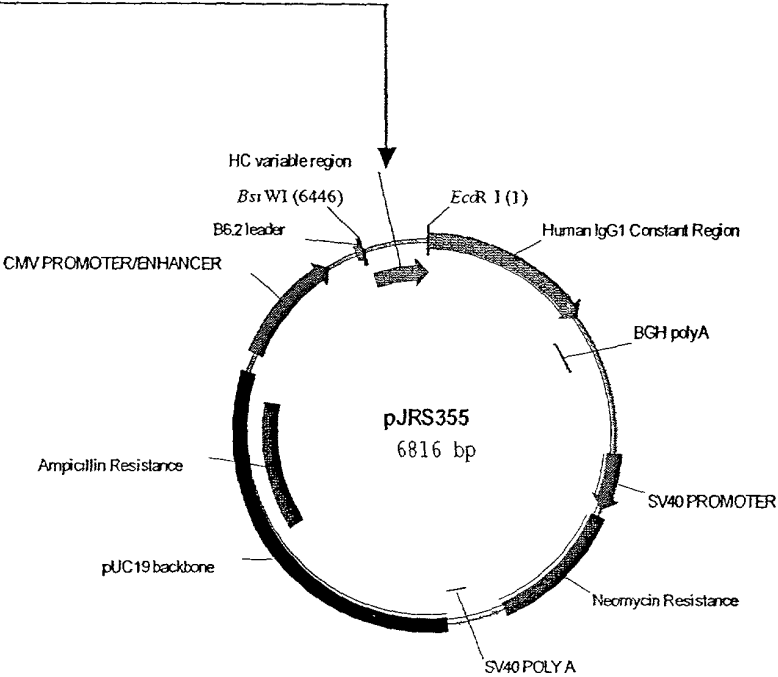
FIG. 8B

Figure A. Human IgG1-cH36 HC Variable Region Cloning and Expression Vector



HC Cloning Vector

Fig. 9A



HC Expression Vector

Fig. 9B

**Figure B. Human IgG4-cH36 HC Variable Region Cloning and Expression Vector**

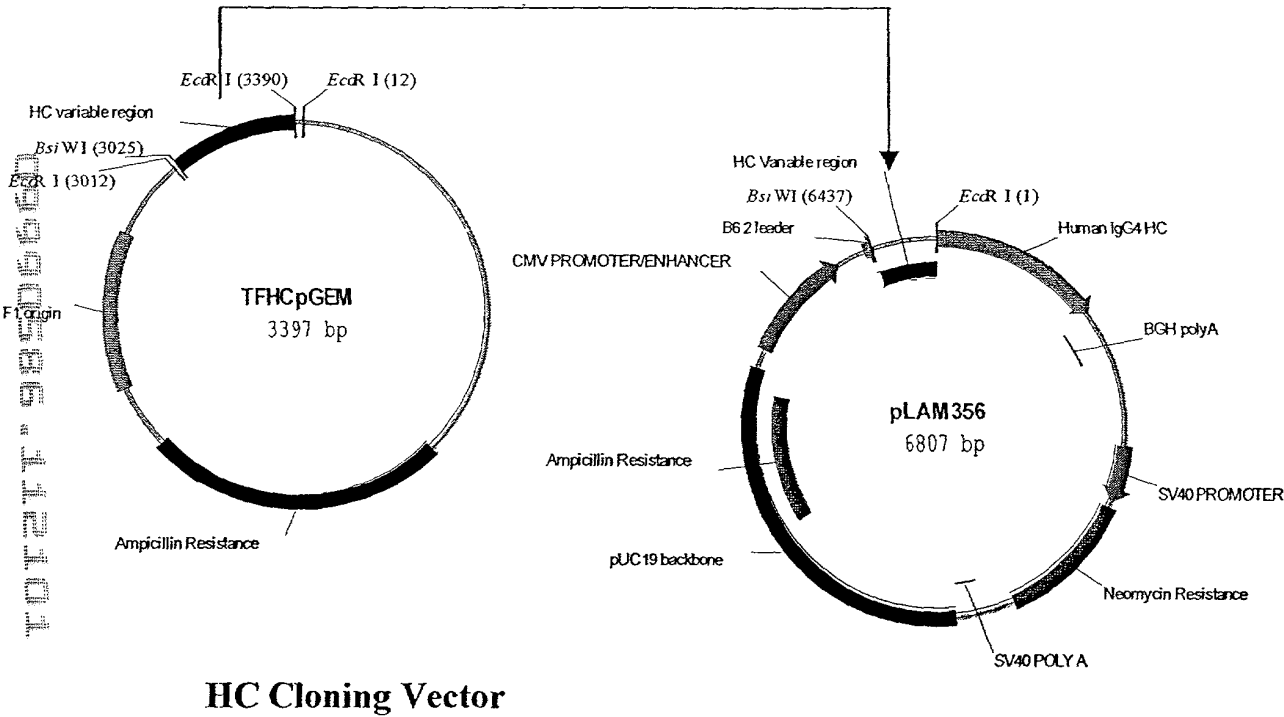


Fig. 9C

**HC Expression Vector**

Fig. 9D

Figure C. cH36 LC Variable Region Cloning and Expression Vector

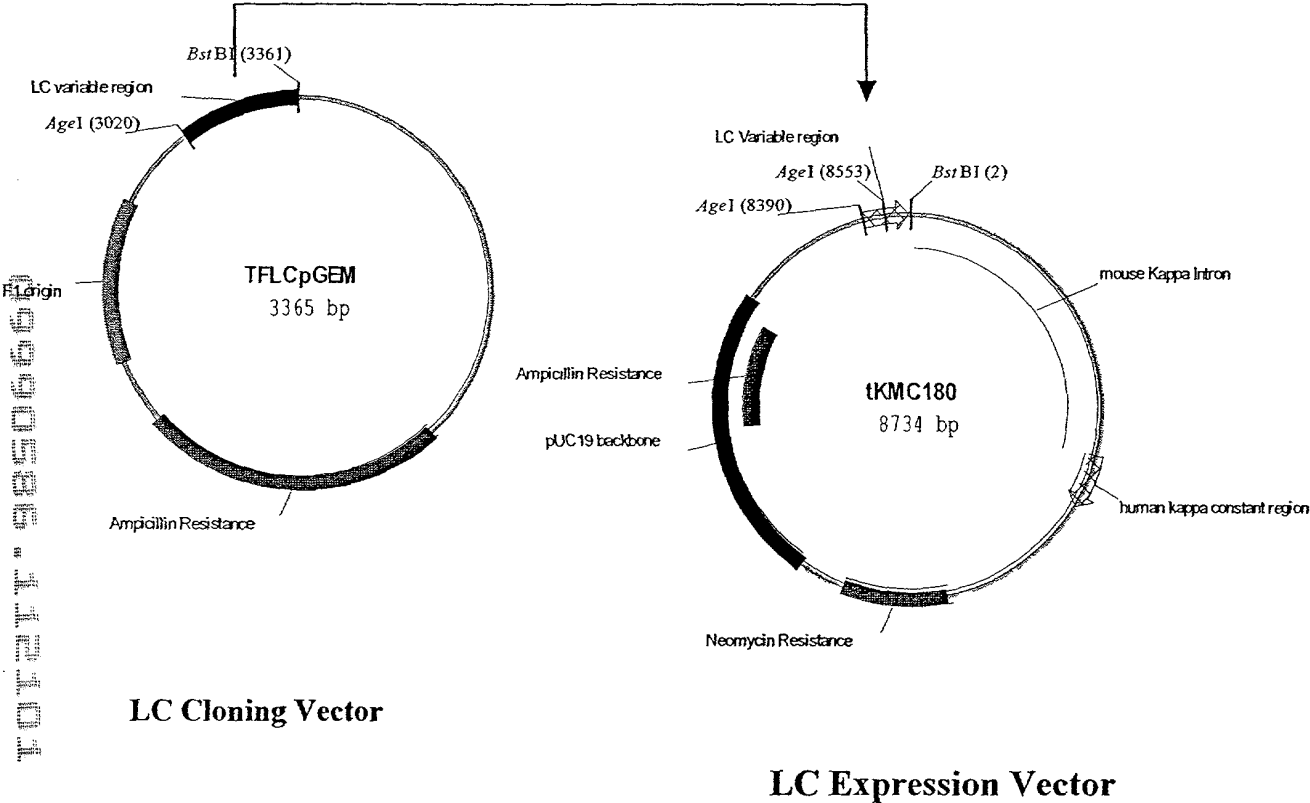


Fig. 10A

Fig. 10B

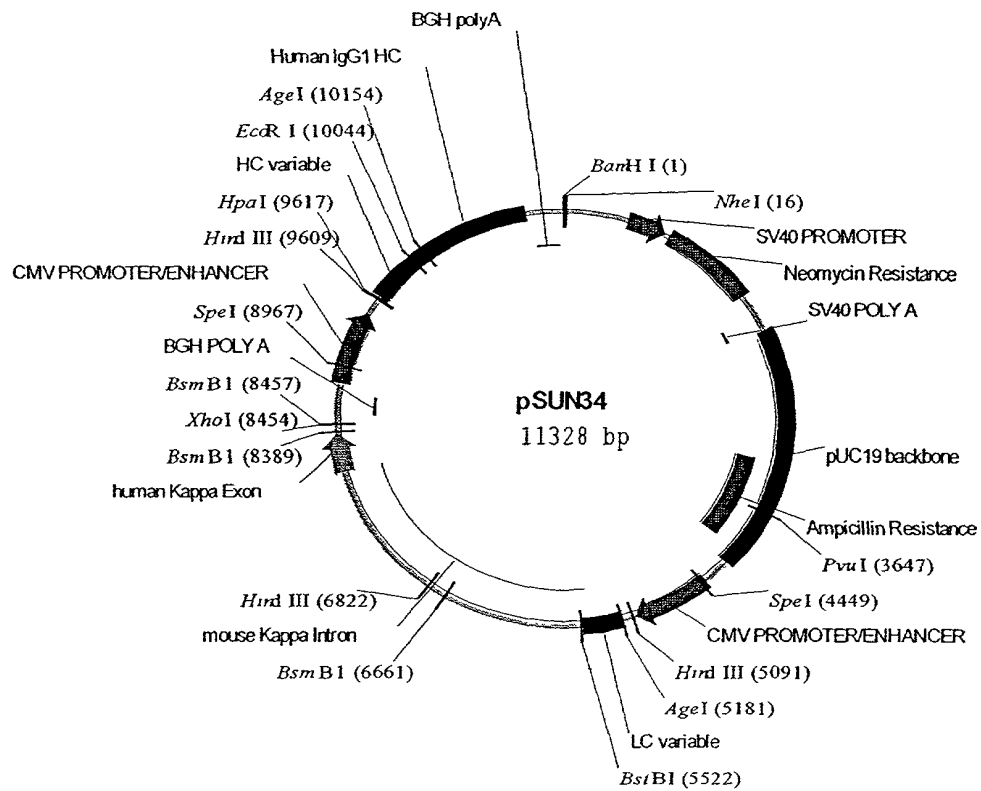


Figure D. Plasmid Map of Humanized Anti-TF IgG1 Antibody Expression Vector

Fig. 11

# Humanization of anti-Tissue Factor Antibody cH36

## Sequences of Partially and Fully Humanized Light Chain (LC) Variable Regions

### Light Chain (LC) FR Sequences

FR1 (23 AA)	FR2 (14 AA)	FR3 (32 AA)	FR4 (10 AA)	Names
1 10 20 35	47 57 60	70	86 98 107	
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	cH36-LC
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-03
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-04
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-05
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-06
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-07
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-08
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-09
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-10
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-11
DIQMTQSPASQASLGE <span style="background-color: #cccccc;">SV</span> TIITC	WYQQKPGKSPQLIY	GVPSRFSGSGSGTKFSFKISSLQAE <span style="background-color: #cccccc;">DFVN</span> YYC	FGAGTKLE <span style="background-color: #cccccc;">LK</span>	LC-12

Fig. 12A

### Light Chain CDR Sequences of cH36

CDR1 (11 AA)	CDR2 (7 AA)	CDR3 (9 AA)
24 34 50 56 89 97		
L A S Q T I D T W L A	A A T N L A D	Q Q V Y S S P F T

Fig. 12B

Fig. 12C

Fig. 12D

# Sequences of Partially and Fully Humanized Heavy Chain (LC) Variable Regions

## Heavy Chain (HC) FR Sequences

FR1 (30 AA)	FR2 (14 AA)	FR3 (32 AA)	FR4 (11 AA)	Names
1 10 20 29 36	44	67 75	85 95 107 117	
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSHGKSLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	CH36-HC
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSHGKSLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-01
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-02
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-03
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-04
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-05
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-06
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-07
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-08
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-08R1
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-11
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-12
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-09
EQIQSQSGPELVKPGASVQVSCCKTSGYSFT	WVRQSGKGLLEWIG	KATLTVDKSSTTAFMHLNSLTSDDSAVYFCAR	WGQGTTLTVSS	HC-10

1-6.  
13A

## Heavy Chain CDR Sequences

CDR1 (5 AA)	CDR2 (17 AA)	CDR3 (8AA)	Names
31 35	50	99 106	
D Y N V Y	Y I D P Y N G I T I Y D Q N F K G	D V T T A L D F	CH36
31 35	50	99 106	
D Y N V Y	Y I D P Y N G I T I Y D Q N F K G	D V T T A L D F	HC-08

Fig. 13C

Fig. 13D

## hOAT (IgG1) Constant regions sequences

### Sequences of LC constant:

RTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKH  
KVYACEVTHQGLSSPVTKSFNRGEC

Fig. 14A

### Sequences of HC constant:

EFASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVTVPSSSLGTQTYIC  
NVNHKPSNTKVDKKVEPKSCDKTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEV  
HNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCL  
VKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFCSCVMHEALHNHYTQKSLSLSPGK

Fig. 14B



# hFAT (IgG4) constant region sequences

## Sequences of LC Constant:

RTVAAPSVFI PPSEQLKSGTASVVC LLNNFY PRAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEK  
HKVYACEVTHQGLSPVTKSFNRGEC

Fig. 15A

## Sequences of HC constant:

EFASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGTQTY  
TCNV D HKPSNTKVDKR VESKYGPPCPCPAPEFLGGPSVFLFPPKPKDTLMISRTPEVTCVVDVVSQEDPEVQFNWYVDGV  
EVHNAKTKPREEQFNSTYRVVSVLTVLHQD WLN GKEYCKVSNKGLPSSIEK TISKAKGQPREPQVYVTLPPSQEEMTKNQVSL  
TCLVKGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSRLTV D KSRWQEGNVFSCSV MHEALHNHYTQKSLSLGLGK

Fig. 15B